Subterranean Biology 28: 1–13 (2018)
doi: 10.3897/subtbiol.28.29282
http://subtbiol.pensoft.net

SHORT COMMUNICATION

A peer-reviewed open-access journal
Subterranean Biology
Published by The International Society for Subterranean Biology

New occurrence records for stygobiontic invertebrates from the Edwards and Trinity aquifers in west-central Texas, USA

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Academic editor: *M. Niemiller* | Received 23 August 2018 | Accepted 30 September 2018 | Published 1 November 2018

http://zoobank.org/AC3F597C-05E8-42A8-A58B-A38B590FCC60

Citation: Nissen BD, Devitt TJ, Bendik NF, Gluesenkamp AG, Gibson R (2018) New occurrence records for stygobiontic invertebrates from the Edwards and Trinity aquifers in west-central Texas, USA. Subterranean Biology 28: 1–13. https://doi.org/10.3897/subtbiol.28.29282

Abstract

We report new occurrence records for stygobiontic invertebrates from the Edwards and Trinity aquifers in Blanco, Hays, and Travis counties of central Texas, USA. Our collection includes seven species from four families: *Caecidotea reddelli* (Steeves, 1968), Asellidae; *Crangonyx* nr. *pseudogracilis* Bousfield, 1958, *Stygobromus balconis* (Hubricht, 1943), *Stygobromus bifurcatus* (Holsinger, 1967), and *Stygobromus russelli* (Holsinger, 1967), Crangonyctidae; *Sphalloplana mohri* Hyman, 1938, Kenkiidae; and *Cirolanides* sp., Cirolanidae. Specimens of *Caecidotea reddelli* and *Crangonyx* nr. *pseudogracilis* are new records for Hays County and Travis county, respectively. Specimens of an undescribed species of *Cirolanides* were collected from a well in Hays County and from two localities in Travis County.

Keywords

karst, groundwater, stygofauna, Asellidae, Crangonyctidae, Kenkiidae, Cirolanidae

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Introduction

The Edwards and Trinity aquifers in west-central Texas are some of the most biologically diverse aquifers in the world, home to at least 68 described species of endemic groundwater-obligate (stygobiontic) invertebrate species (Hershler and Longley 1986, Bowles and Arsuffi 1993, Hutchins 2018, Külköylüoğlu et al. 2017a, 2017b, 2017c, Camacho et al. 2018, Külköylüoğlu 2018, Külköylüoğlu and Gibson 2018). Of these species, 52 are in the Edwards (Balcones Fault Zone) Aquifer (hereafter, Edwards Aquifer), 18 are in the Trinity Aquifer, and 23 are in the Edwards-Trinity (Plateau) Aquifer. Seventeen stygobiontic invertebrates have been recorded in the Austin-Round Rock Metro Area (Table 1). The Edwards Aquifer is also the primary source of water for the city of San Antonio and other communities in central Texas (Gibson et al. 2008), which are currently experiencing rapid development and growth (Pendall et al. 2015). As new pumping wells are drilled for agricultural and municipal use, increased demands are placed on these aquifers, threatening regional groundwater biodiversity and ecosystem services. Documenting the stygobionts endemic to these aquifers is necessary to refine our knowledge of their distributions, thereby informing conservation and management of natural resources within the Edwards and Trinity aquifers. Furthermore, monitoring stygobiontic communities can prove useful in detecting changes in the water quality of these aquifers (Gibson et al. 2008), which many Central Texans rely on for drinking water, agriculture, and recreation.

Despite considerable recent research (Bowles and Arsuffi 1993, Gibson et al. 2008, Diaz and Alexander 2010, Hutchins et al. 2013), the distributions of stygobionts remain difficult to delineate due to the inaccessibility of their habitats, and low detection probabilities (Schneider and Culver 2004, Krejca and Weckerly 2007). Here, we

Table 1. Stygobiontic invertebrate fauna recorded in the Austin-Round Rock Metro area, Texas, USA.

Class	Order	Family	Species
Turbellaria	Kenkiidae	Kenkiidae	*Sphalloplana mohri* Hyman, 1938
Mollusca	Mesogastropoda	Hydrobiidae	*Phreatodrobia conica* Hershler & Longley, 1986
			Phreatodrobia nugax (Pilsbry & Ferriss, 1906)
			Phreatodrobia punctata Hershler & Longley, 1986
			Phreatodrobia rotunda Hershler & Longley, 1986
		Cochliopidae	*Stygopyrgus bartonensis* Hershler & Longley, 1986
Crustacea	Isopoda	Asellidae	*Caecidotea reddelli* (Steeves, 1968)
			Lirceolus bisetus (Steeves, 1968)
			Lirceolus hardeni (Lewis & Bowman, 1996)
		Cirolanidae	*Cirolanides texensis* Benedict, 1896
	Amphipoda	Bogidiellidae	*Artesia subterranea* Holsinger, 1980
		Crangonyctidae	*Crangonyx* nr. *pseudogracilis* Bousfield, 1958
			Stygobromus balconis (Hubrict, 1943)
			Stygobromus bifurcatus (Holsinger, 1967)
			Stygobromus flagellatus (Benedict, 1896)
			Stygobromus russelli (Holsinger, 1967)
		Sebidae	*Seborgia relicta* Holsinger, 1980

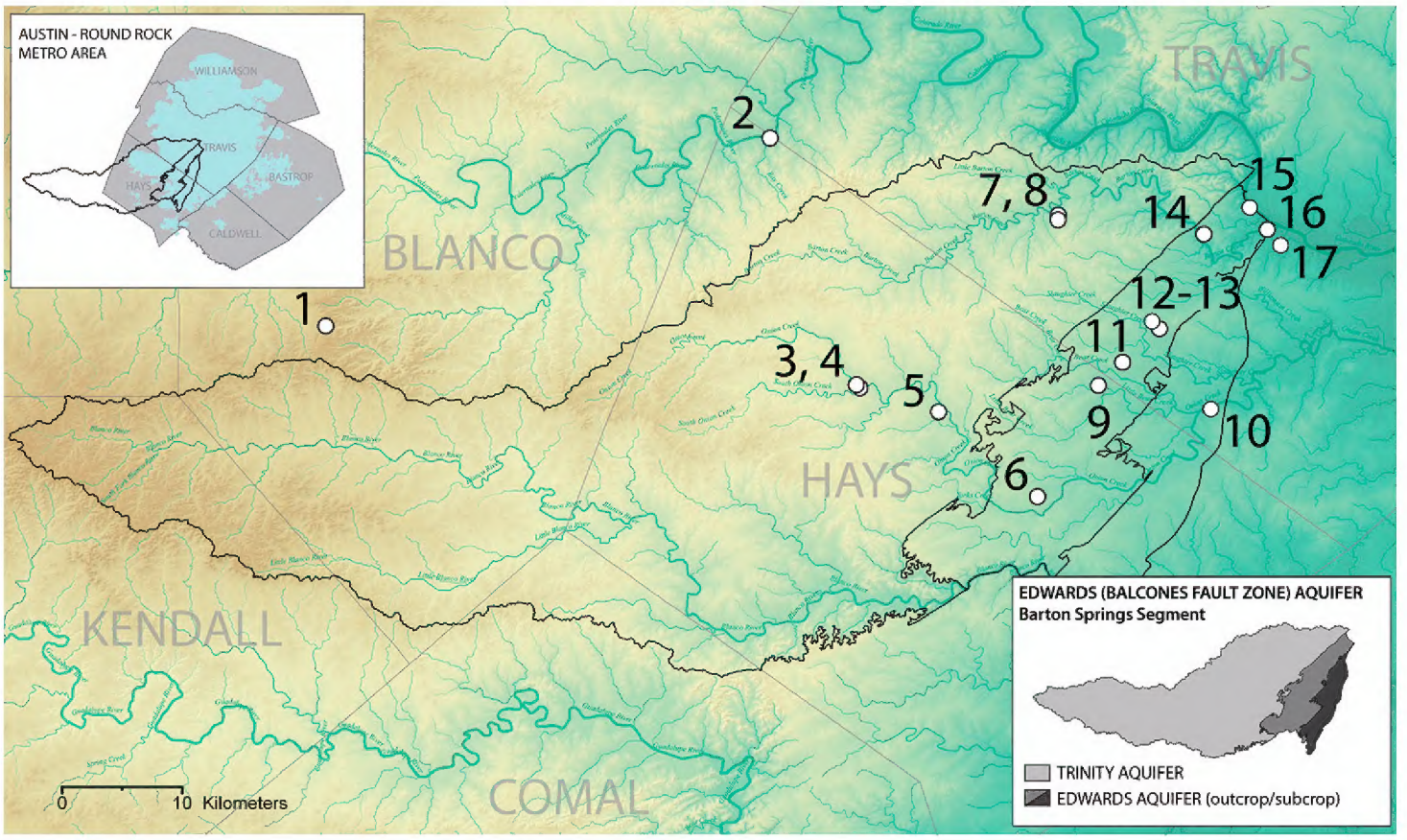

Figure 1. Sample Sites. Sampling map showing the extent of the Barton Springs Segment of the Edwards (Balcones Fault Zone) Aquifer and its hydrozones in Hays, Travis, and Blanco counties, Texas, USA. Sampling sites are numbered as follows: **1** Bamberger Ranch Spring **2** Red's Spring **3** Emerald Spring **4** Bello Spring **5** Ben McCulloch Spring **6** Sky Ranch Tract - State Well No. 5857507 **7** Sweetwater Spring 4 **8** Sweetwater Spring 1 **9** Hays County Ranch Tract - State Well No. 5849939 **10** Old San Antonio Spring **11** Ed's Crossing Tract - State Well No. 58499SH **12** Blowing Sink Cave **13** Blowing Sink Tract - State Well No. 5850411 **14** Barton Creek Greenbelt - State Well No. 5842820 **15** Cold Spring **16** Eliza Spring **17** Treadwell Spring. Boundaries of aquifer hydrozones courtesy of the Barton Springs Edwards Aquifer Conservation District. Wells are identified primarily by the Texas Water Development Board (TWDB) well-numbering system (Nordstrom and Quincy 1999).

present new occurrence records for seven species, including three new county records, from groundwater wells and springs in the Edwards and Trinity aquifers. Long-term monitoring of groundwater wells using bottle-traps allows a unique opportunity to sample a variety of locations over long periods of time with minimal effort (Hutchins and Orndorff 2009, Fenolio et al. 2017). In addition, we give a brief synopsis of known distributions of those species and relevant literature.

Methods

Sampling sites

Seventeen sites were sampled in and around the Barton Springs segment of the Edwards Aquifer and its catchment area in the Hill Country portion of the Trinity Aquifer in Blanco, Hays, and Travis counties (Figure 1). Most sampling took place in 2010–2011 and again in 2015–2018. We also report a handful of other specimens collected opportunistically over the past two decades (Table 2).

Table 2. Voucher specimens. Complete listing of all specimens collected. UTIC = University of Texas Insect Collection. Collector initials are as follows: TJD = Thomas J. Devitt; BDN = Bradley D. Nissen; MSS = Mark S. Sanders; NFB = Nathan F. Bendik; AGG = Andrew G. Gluesenkamp; RG = Randy Gibson; DAC = Dee Ann Chamberlain; PS = Peter Sprouse. N = Specimens collected. † = new county record. * = specimen accessioned at San Marcos US Fish and Wildlife Service Fish Hatchery.

Taxon	Sites	N	Date	Collectors	Catalog #
Caecidotea reddelli	Hays Co.: Roy Creek, Red's Spring†	4	16 Sep 2016	TJD	UTIC 92016
	Travis Co.: Zilker Park, Eliza Spring	1	1 Apr 1999	DAC	UTIC 93008
	Travis Co.: Barton Creek Habitat Preserve, Sweetwater Spring 4	4	10 Apr 2017	TJD, BDN	UTIC 92021
		3	17 Apr 2017	TJD, BDN	UTIC 92020
	Travis Co.: Barton Creek Habitat Preserve, Sweetwater Spring 1	3	17 Apr 2017	TJD, BDN	UTIC 92019
		1	1 May 2017	TJD, BDN	UTIC 92018
	Travis Co.: Old San Antonio District Park, Old San Antonio Spring	2	19 Jan 2018	TJD, BDN	UTIC 93014
Cirolanides nr. *texensis*	Travis Co.: Blowing Sink Cave†	1	14 Oct 2010	MSS	UTIC 91874
	Travis Co.: City of Austin WQPL, Blowing Sink Tract, State Well No. 5850411	1	4 Dec 2017	BDN	UTIC 210886
	Hays Co.: City of Austin WQPL, Hays County Ranch Tract, State Well No. 5849939†	2	1 Sep 2010	NFB, AGG	UTIC 91879
		3	12 Nov 2010	NFB, AGG	UTIC 91876
		1	3 Dec 2010	NFB, AGG	UTIC 91877
		3	14 Jan 2011	NFB, AGG	UTIC 91875
		2	27 Jan 2011	NFB, AGG	UTIC 91880
		2	21 Oct 2016	AGG, TJD, BDN	UTIC 92014
		1	15 Nov 2016	BDN	UTIC 92013
		3	6 Apr 2017	BDN	UTIC 210884
		2	20 Apr 2017	BDN	UTIC 210885
		1	5 Jan 2018	BDN	UTIC 210887
Crangonyx nr. *pseudogracilis*	Hays Co.: Old San Antonio Spring	2	31 Jan 2018	BDN	Cp31012018*
	Travis Co.: Treadwell Spring	3	21 June 2016	PS	UTIC 91369
Sphalloplana mohri	Travis Co.: Cold Spring	1	24 Feb 2011	RG	SM-Sm24022011*
Stygobromus balconis	Travis Co.: City of Austin WQPL, Ed's Crossing Tract, State Well No. 58499SH	1	6 Apr 2017	BDN	UTIC 92024
	Travis Co.: Barton Creek Habitat Preserve, Sweetwater Spring 4	4	10 Apr 2017	BDN	UTIC 92025
Stygobromus bifurcatus	Travis Co.: Zilker Park, Eliza Spring	1	29 Aug 2016	DAC	UTIC 92030
		1	5 Mar 2017	DAC	UTIC 93011
	Travis Co.: Barton Creek Habitat Preserve, Sweetwater Spring 4	8	17 Apr 2017	TJD, BDN	UTIC 92026
	Hays Co.: Onion Creek, Ben McCulloch Spring	1	31 Jan 2017	TJD	UTIC 92029
	Blanco Co.: Bamberger Ranch Spring	1	21 Jun 2017	TJD, BDN	UTIC 92028
Stygobromus russelli	Blanco Co.: Bamberger Ranch Spring	1	22 Mar 2018	TJD, BDN	UTIC 93016
	Travis Co.: Zilker Park, Eliza Spring	1	19 Nov 2015	DAC	UTIC 92033
	Travis Co.: Barton Creek Wilderness Park, Barton Creek Greenbelt tract, State Well No. 5842820	3	3 Dec 2010	NFB, AGG	UTIC 91888
		2	27 Jan 2011	NFB, AGG	UTIC 91882
		1	8 Mar 2011	NFB, AGG	UTIC 91883
	Travis Co.: City of Austin WQPL, Ed's Crossing Tract, State Well No. 58499SH	2	8 Mar 2011	NFB, AGG	UTIC 91886
		2	4 Dec 2017	BDN	UTIC 93012
		1	3 Jan 2018	BDN	UTIC 93013
	Travis Co.: City of Austin WQPL, Blowing Sink Tract, State Well No. 5850411	1	30 Mar 2018	BDN	UTIC 93017

Taxon	Sites	N	Date	Collectors	Catalog #
Stygobromus russelli	Hays Co.: Onion Creek, Ben McCulloch Spring	1	31 Jan 2017	TJD	UTIC 92029
		3	21 Mar 2017	TJD, BDN	UTIC 92031
		5	3 May 2017	TJD, BDN	UTIC 92039
	Hays Co.: City of Austin WQPL, Sky Ranch Tract, State Well No. 5857507	1	1 Sep 2010	NFB, AGG	UTIC 91889
		3	3 Dec 2010	NFB, AGG	UTIC 91885
		8	14 Jan 2011	NFB, AGG	UTIC 91887
		1	8 Mar 2011	NFB, AGG	UTIC 91884
	Hays Co.: Onion Creek, Bello Spring	2	18 Apr 2017	TJD, BDN	UTIC 92035
	Hays Co.: South Onion Creek, Emerald Spring	1	13 Jan 2017	TJD, BDN	UTIC 92040
		2	18 Apr 2017	TJD, BDN	UTIC 92032
	Hays Co.: Roy Creek, Red's Spring	2	23 Apr 2017	TJD, BDN	UTIC 92038
		1	25 Jun 2017	TJD, BDN	UTIC 92037

Trap designs

We sampled springs using cotton mophead "traps" lodged into spring outlets (modified per methods in Holsinger and Minckley 1971, Hershler and Longley 1986, Gibson et al. 2008, Huston et al. 2015). We separated the mopheads into individual strings, tied them into loose bunches, and securely wedged them into the spring outlets using rocks to keep them in place (Figure 2). The size of the mop bunches was determined by the size of the spring outlet, ideally filling a large portion of the outlet, to maximize the volume of water flowing through the trap. Where possible, we placed multiple clumps of mophead material into the spring outlets at various locations. We checked mops for invertebrates after approximately two weeks by removing them from the spring outlet, quickly placing them in a large handheld net and flushing water through the net to dislodge any invertebrates from mophead strands. We also searched through the strands by hand after flushing. Specimens were collected and stored in 99% ethanol.

Five groundwater wells were sampled using a funnel trap fashioned from 1-L plastic water bottles with the top cut off and inverted into the bottle (Fenolio et al. 2017). We baited the traps using pistachio nuts, dried Mysis shrimp, Slim Jim (Conagra Brands) pieces, or catfish bait (Catalpa Worm: Little Stinker (Acme Tackle Company); Cricket: Berkley Gulp! Alive! (Berkley Fishing)). Traps were set between 0–10 m above the bottom of the well (Table 3). Wells were sampled every two weeks from September 2010

Table 3. Well Information. Data on sampled wells collected in the field and from the TWDB database. Tract names correspond to City of Austin WQPL tracts. The depth of trap in State Well No. 5857507 (Sky Ranch Tract) was not recorded. Water depth measurements were taken in the fall of 2017.

Tract Name/State Well No.	Depth to water	Depth of trap	Depth to bottom of well
Hays Co. Ranch – 58-49-939	17.2 m	27.7 m	29.3 m
Ed's Crossing – 58-49-9SH	42.6 m	44.2 m	44.2 m
Blowing Sink – 58-50-411	69.1 m	84.4 m	96.0 m
Barton Creek – 58-42-820	80.1 m	unknown	137 m
Sky Ranch – 58-57-507	48.8 m	unknown	306 m

Figure 2. Mophead in spring outlet at Cold Spring, Travis County, Texas, USA.

to December 2011 and then again 22 times between 21 October 2016 and 30 March 2018, for a total of over 1,000 days of trapping effort. Specimens are deposited in the Biodiversity Collections of the University of Texas at Austin and at the San Marcos Aquatic Resources Center, United States Fish and Wildlife Service (Table 2).

Results

New Occurrence Records

Caecidotea reddelli (Steeves, 1968) (Isopoda, Asellidae)

Site 1. TEXAS: Hays County: Roy Creek: Red's Spring (30.36324, -98.12315). Two specimens collected 21 October 2016 by TJD. Identified by RG. *New county record.*

Site 2. TEXAS: Travis County: Zilker Park, Eliza Spring (30.26425, -97.77006). One specimen collected 1 April 1999 by Dee Ann Chamberlain. Identified by RG.

Site 3. TEXAS: Travis County: Barton Creek Habitat Preserve, Sweetwater Tract Spring 1 (30.27535, -97.92709). One specimen collected 1 May 2017 by BDN and TJD. Identified by RG.

Site 4. TEXAS: Travis County: Barton Creek Habitat Preserve, Sweetwater Tract Spring 4 (30.27171, -97.92731). Three specimens collected 17 April 2017 by BDN and TJD. Identified by RG.

Site 5. TEXAS: Travis County: Old San Antonio District Park, Old San Antonio Spring (30.13217, -97.81750). Two specimens collected 19 January 2018 by BDN and TJD. Identified by RG.

Caecidotea reddelli is a stygobiontic isopod about 10 mm in length that occurs throughout central Texas in springs, caves, and wells in Bell, Burnet, Coryell, Dallas, Henderson, Hill, Limestone, Palo Pinto, Panola, San Augustine, Tarrant, Travis, and Williamson counties (Steeves 1968, Mitchell and Reddell 1971, Lewis and Bowman 1996, Lewis 2001, Hutchins 2018). *Caecidotea reddelli* is known from both the North Balcones Fault Zone and the adjacent part of the Gulf Coast Plain Province directly to the northeast in Dallas and Henderson counties (Lewis and Bowman 1996). Mitchell and Reddell (1971) showed an additional locality in Hays County (their fig. 30) but without further explanation, and there is no associated voucher specimen (Reddell, personal communication to TJD and BDN, 21 September 2017). Therefore, we present our specimen of *C. reddelli* collected from Red's Spring in Hays County as a new county record for this species.

***Cirolanides* nr. *texensis* (Isopoda, Cirolanidae).**

Site 1. TEXAS: Hays County: City of Austin Water Quality Protection Lands (WQPL), Hays County Ranch Tract, State Well No. 5849939 (30.14722, -97.89691). First two specimens collected 1 September 2010 by NFB and AGG. Additional specimens are listed in Table 2. All specimens were identified by Benjamin F. Schwartz (Texas State University).

Site 2. TEXAS: Travis County: Blowing Sink Cave (30.189718, -97.851014). One specimen collected 14 October 2010 by Mark S. Sanders. Identified by Benjamin F. Schwartz. *New county record.*

Site 3. TEXAS: Travis County: City of Austin WQPL, Blowing Sink Tract, State Well No. 5850411 (30.18667, -97.84917). One specimen collected 4 December 2017 by BDN. Identified by Benjamin F. Schwartz.

These specimens are part of the *Cirolanides texensis* species complex, which needs revision (Ben Hutchins, Texas Parks and Wildlife, personal communication to BDN on 12 July 2018). These specimens represent a distinct lineage of *Cirolanides* related to *C. texensis* (Benedict, 1896) that warrants species-level designation, to be described elsewhere (Benjamin F. Schwartz, Texas State University, personal communication to BDN on 15 December 2017). All specimens were collected from a Hays County well and a cave and nearby well in Travis County.

***Crangonyx* nr. *pseudogracilis* (Bousfield, 1958) (Amphipoda, Crangonyctidae)**

Site 1. TEXAS: Hays County: Old San Antonio District Park, Old San Antonio Spring (30.13217, -97.8175). Two specimens collected 31 January 2018 by BDN. Identified by RG.

Site 2. TEXAS: Travis County: Treadwell Spring (30.2549698, -97.7592774). Three specimens collected 21 June 2016 by Peter Sprouse. Identified by RG. *New county record.*

Crangonyx pseudogracilis is recorded in the east-central United States and southern Canada (Zhang and Holsinger 2003). Diaz and Alexander (2010) noted specimens of *Crangonyx* sp. collected in samples from the spring-fed San Marcos River, Hays County, Texas. Groundwater-adapted populations with reduced eyes and reduced pigmentation have been recorded in Comal and Kendall counties (Gibson et al. 2008). Specimens collected from Old San Antonio Spring and Treadwell Spring also show these stygobiontic adaptations.

***Sphalloplana mohri* (Hyman, 1938) (Triclada, Kenkiidae)**

Site 1. TEXAS: Travis County: Cold Spring (30.27959, -97.78043). One specimen collected 24 February 2011 by RG. Identified by RG.

Sphalloplana mohri is a relatively large flatworm described by Hyman (1938) and recorded from the Edwards Plateau in Hays, Kendall, Mason, San Saba, Travis, and Uvalde counties (Kenk 1977, Hutchins 2018). This is the first record for Cold Spring.

***Stygobromus balconis* (Hubricht, 1943) (Amphipoda, Crangonyctidae)**

Site 1. TEXAS: Travis County: City of Austin WQPL, Ed's Crossing Tract, State Well No. 58499SH (30.16472, -97.87889). One specimen collected 6 April 2017 by BDN. Identified by RG.

Site 2. TEXAS: Travis County: Barton Creek Habitat Preserve, Sweetwater Tract Spring 4 (30.27171, -97.92731). Four specimens collected 10 April 2017 by TJD and BDN. Identified by RG.

Stygobromus balconis is a relatively large species of *Stygobromus*. This species was originally described by Hubricht (1943) and later redescribed by Holsinger (1966, 1967) who subdivided the taxon into three species: *S. russelli*, *S. bifurcatus,* and *S. balconis*. *Stygobromus balconis* is known from very few localities in Hays, Travis and Kendall counties (Hutchins 2018).

***Stygobromus bifurcatus* (Holsinger, 1967) (Amphipoda, Crangonyctidae)**

Site 1. TEXAS: Travis County: Zilker Park, Eliza Spring (30.26425, -97.77006). One specimen collected 29 August 2016 by Dee Ann Chamberlain. Identified by RG.

Site 2. TEXAS: Travis County: Barton Creek Habitat Preserve, Sweetwater Tract Spring 4 (30.27171, -97.92731). Eight specimens collected 17 April 2017 by TJD and BDN. Identified by RG.

Site 3. TEXAS: Hays County: Onion Creek, Ben McCulloch Spring (30.12732, -98.01709). One specimen collected 31 January 2017 by TJD. Identified by RG.

Site 4. TEXAS: Blanco County: Bamberger Ranch Spring (30.19185, -98.47723). One specimen collected 21 June 2017 by TJD and BDN. Identified by RG.

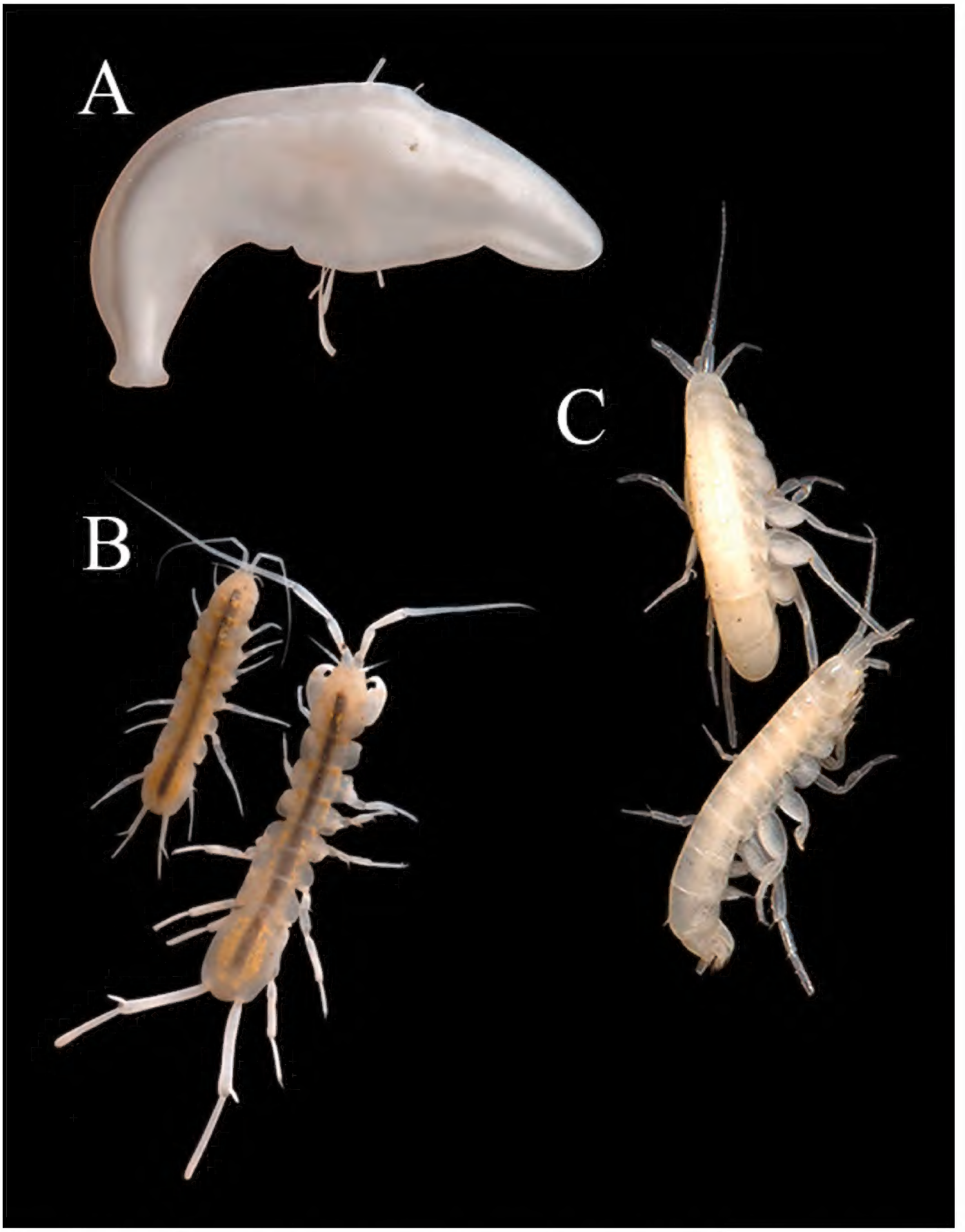

Figure 3. A *Sphalloplana mohri* Hyman, 1938 from Cold Spring, Travis Co., Texas, USA **B** *Caecidotea reddelli* (Steeves, 1968) from Rocket River Cave, Coryell Co., Texas, USA **C** *Stygobromus balconis* (Hubricht, 1943) from Autumn Woods Well, Hays Co., Texas, USA. All photographs by Dr. Jean K. Krejca, Zara Environmental LLC. Images not to scale.

Stygobromus bifurcatus (Holsinger, 1967) is rather widely distributed, often occurring syntopically with *S. russelli* (Mitchell & Reddell, 1971). It is currently known from Bell, Bexar, Blanco, Burnet, Comal, Coryell, Hays, Kendall, Lampasas, San Saba, Travis, and Williamson counties (Hutchins 2018).

***Stygobromus russelli* (Holsinger, 1967) (Amphipoda, Crangonyctidae)**

Site 1. TEXAS: Travis County: Barton Creek Greenbelt, State Well No. 5842820 (30.26139, -97.816944). Three specimens first collected 3 December 2010 by AGG and NFB. Additional specimens listed in Table 2. All specimens identified by RG.

Site 2. TEXAS: Travis County: Zilker Park, Eliza Spring (30.26425, -97.77006). One specimen collected 19 November 2015 by Dee Ann Chamberlain. Identified by RG.

Site 3. TEXAS: Travis County: City of Austin WQPL, Blowing Sink Tract, State Well No. 5850411 (30.18667, -97.84917). One specimen collected 30 March 2018 by BDN. Identified by RG.

Site 4. TEXAS: Hays County: South Onion Creek, Emerald Spring (30.14769, -98.07868). One specimen collected 13 January 2017 by TJD and BDN. Additional specimens listed in Table 2. Identified by RG.

Site 5. TEXAS: Travis County: City of Austin WQPL, Ed's Crossing Tract, State Well No. 58499SH (30.16472, -97.87889). One specimen collected 8 Mar 2011 by AGG and NFB. Additional specimens listed in Table 2. Identified by RG.

Site 6. TEXAS: Hays County: Onion Creek, Bello Spring (30.14537, -98.07599). Two specimens collected 18 April 2017 by TJD and BDN. Identified by RG.

Site 7. TEXAS: Hays County: Onion Creek, Ben McCulloch Spring (30.12732, -98.01709). One specimen first collected 31 January 2017 by TJD. Additional specimens listed in Table 2. All specimens were identified by RG.

Site 8. TEXAS: Hays County: City of Austin WQPL, Sky Ranch Tract, State Well No. 5857507 (30.06358, -97.94253). One specimen first collected 1 September 2010 by AGG and NFB. Additional specimens listed in Table 2. All specimens identified by RG.

Site 9. TEXAS: Hays County: Roy Creek, Red's Spring (30.36324, -98.12315). Two individuals first collected 23 April 2017 by TJD. Additional specimens are listed in Table 2. All specimens were identified by RG.

Site 10. TEXAS: Blanco County: Bamberger Ranch Spring (30.19185, -98.47723). One specimen collected 22 March 2018 by TJD and BDN. Identified by RG.

Stygobromus russelli is relatively common, morphologically variable, and widely distributed throughout the Edwards and adjacent Trinity aquifers (Hutchins et al. 2013). Its range covers most of the eastern half of the limestone area of central Texas, however most records are recorded from caves just west and northwest of Austin (Holsinger and Longley 1980). It is currently known from Bandera, Bell, Bexar, Burnet, Comal, Coryell, Hays, Kendall, Kerr, Mason, Medina, San Saba, Travis, and Williamson counties (Hutchins 2018).

Acknowledgments

We thank James Reddell (University of Texas) for cataloging all specimens, and valuable feedback; Benjamin Schwartz (Texas State University) and Ben Hutchins (Texas Parks and Wildlife Department) for specimen identification and helpful discussion; and Dee Ann Chamberlain (City of Austin), Mark Sanders (City of Austin), and Pe-

ter Sprouse (Zara Environmental, LLC) for specimen donations. Finally, we thank the many landowners and managers who provided access to lands under their care, including: Jared Holmes and the Bamberger Ranch; Reed and Holton Burns (Charro Ranch); Kevin Theusen (City of Austin); the owners of Camp Ben McCulloch; Brandon Crawford, John Karges, and The Nature Conservancy of Texas. The views presented herein are those of the authors and do not necessarily represent those of the U.S. Fish and Wildlife Service or the City of Austin.

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